Atovaquone for the Treatment of COVID-19: ATaQ

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Atovaquone for Treatment of COVID-19

Ataq COVID-19

Study Design: This is a randomized, double-blind study of atovaquone therapy in adult participants hospitalized with COVID-19. Approximately 60 participants who meet all eligibility criteria may be randomized in a 2:1 atovaquone/placebo ratio into one of the following treatment groups:

Treatment Group 1: continued standard of care therapy together with an oral dose of 1500 mg atovaquone twice daily (administered with a meal or snack) for up to 10 days

Treatment Group 2: continued standard of care therapy together with matching placebo

Primary Efficacy Endpoint: Viral load (Log copy number/ml by RT-PCR) from saliva collection at Day 8 from trial entry, or at hospital discharge (if discharged before Day 8).

Secondary Efficacy Endpoints:

1. Change in viral load (Log copy number/ml) from baseline to Day 8 (or to discharge if discharged before day 8).
2. Viral load (log copy number/ml by RT/PCR) from saliva collection at Day 15 or hospital discharge if discharged prior to Day 15.
3. Days from baseline to decrease in viral load by ≥2 Log copy number/ml.
4. Proportion with viral clearance (undetectable by RT-PCR) by last day of hospitalization or Day 8 of trial.
5. Proportion with viral clearance (undetectable by RT-PCR) by last day of hospitalization or Day 15 of trial.
6. Proportion with ≥2 Log copy number/ml change by Day 8 or by discharge from hospital if discharged before Day 8.
7. Proportion with ≥2 Log copy number/ml change by Day 15 or by discharge from hospital if discharged before Day 15.
8. Days to viral clearance among those with viral clearance (undetectable by RT-PCR) on last day of hospitalization or Day 8 if still hospitalized.
9. Days to viral clearance among those with viral clearance (undetectable by RT-PCR) on the last day of hospitalization of Day 15 if still hospitalized.
10. Exploratory clinical endpoint: Time to clinical improvement of ≥2 points on the ordinal clinical scale or discharge alive, whichever comes first
11. Exploratory endpoint: Change in routinely collected clinical biomarkers such as CRP, ferritin, D-Dimer, IL-6 hs-cTnT and NT-proBNP among those randomized to treatment vs. placebo from baseline through day 8.

For all analyses of viral load and clearance, results from salvia samples will be used for primary and secondary analyses. If salvia cannot be obtained (i.e due to mechanical ventilation), a nasopharyngeal swab can be used.

Ordinal Clinical Scale

The ordinal scale is an assessment of the clinical status at the first assessment of a given study day. Obtain Day 8 Ordinal scale

1. Death
2. Hospitalized, on invasive mechanical ventilation or ECMO
3. Hospitalized, on non-invasive ventilation or high flow oxygen devices
4. Hospitalized, requiring supplemental oxygen
5. Hospitalized, not requiring supplemental oxygen
6. Discharged

Safety Endpoint: Between group difference in treatment-emergent adverse events leading to study drug dose reduction or discontinuation.

Statistical Methods:

All analyses will be performed among randomized participants who received at least one dose of study drug and has at least one post-baseline viral load measurement.

1. Given the skewed distribution of the viral load measurement, viral load will be compared at Day 8 and end of hospitalization among those who are discharged prior to Day 8 between groups with the Wilcoxon rank-sum test.
2. Differences between baseline and day 8 or last day of hospitalization if discharged prior to Day 8 viral load will be compared between groups with the Wilcoxon rank-sum test. Within group differences will be assessed via the Wilcoxon signed-rank test.
3. Days from baseline to decrease in viral load by 2 log copy number/ml will be determined using Kaplan-Meier analysis, and the treatment groups will be compared with the log-rank test. The median number of days from baseline to the decrease in viral load will be compared via the Wilcoxon rank-sum test.
4. Proportions with viral clearance ≥ 2 log copy number change/mL at Day 8 or last day of hospitalization if discharged prior to Day 8 will be compared between groups with the Chi square test.
5. Days to clinical improvement ≥2 points on the ordinal scale or discharged alive, whichever comes first, will be determined using Kaplan-Meier analysis, and the treatment groups will be compared with the log-rank test. The change in ordinal score will be compared between groups via the Wilcoxon rank-sum test.

There is no methodological approach for handling missing data values that is universally accepted in all situations. However, we propose to use an iterative Markov chain Monte Carlo method that can be used when the pattern of missing data is monotone (i.e. a

patient attends all visits until a visit is missed and never returns) or non-monotone. Last observation carried forward (LOCF) is generally considered as a conservative approach, but the MCMC simulation method we propose will reduce the risk of overestimation of the precision of the treatment effect.

Sample Size and Statistical Power:

A total of 60 participants will be randomized in a 2:1 ratio to 2 groups (Approximately 40 participants in atovaquone group and 20 in placebo group) This sample size would result in the following statistical power for the viral clearance endpoints

1. For the endpoint of viral load (log copy number/ml) at Day 8 and last day of hospitalization [1], assuming a mean difference between the groups of 1.6 log copy number/mL, with equal standard deviations of 2, the power will be 82%
2. For the endpoints of differences between baseline and Day 8 or last day of hospitalization viral load, assuming, a mean difference-in-difference of >2.5 with a common SD of 3, the power will be >80%
3. For the endpoint of days from baseline to decrease in virial load by ≥2 log copy number/ml, the power will be >90% assuming a difference of 2 days between the treatment groups with a standard deviation of 1 day.
4. For the analyses of the proportions with viral clearance, and the proportion with ≥2 log copy number change at Day 8, the study will have > 80% power to detect a 40% absolute difference between groups and 62% power to detect a 30% difference between groups.
5. Because the ordinal scale and biomarker comparisons are exploratory, no power calculations are provided

Aggregate blinded viral load data will be reviewed during the study and if the magnitude and/or rate of viral clearance is lower than estimated, or the standard deviation is higher, consideration will be given to increase the sample size accordingly.

Interim Analysis:

The Data Safety and Monitoring Board (DSMB) will meet to evaluate data when 50% of enrolled patients have completed 10 days. The DSMB will review number of deaths, number of those on ventilators, and grade 3 and 4 drug related toxicities.

Background

In December 2019, a series of pneumonia cases of unknown cause emerged in Wuhan, Hubei, China. Sequencing analysis from the patients' respiratory tract samples indicated a novel coronavirus (CoV), which was named COVID-19. As of April 21 2020, more than 9 million confirmed cases have been identified globally. More than 400,000 deaths associated with COVID-19 have been reported, making COVID-19 and major health emergency. On March 11, World Health Organization (WHO) declared the resulting disease named COVID-19 as a pandemic[2]. Clinical efforts to discover potential vaccines and therapeutics are still ongoing with no clear treatment or prophylaxis for COVID-19 in sight. It is safe to say that a sufficient understanding of SARSCoV-2, and the full clinical picture of the resulting COVID-19 disease will take some time. Similarly, developing and

widely distributing effective vaccines or novel antiviral drugs is unlikely to occur during this season, which leaves healthcare systems vulnerable, and risks high mortality rates. Alternatively, drug repurposing strategies can create viable pathways towards identification of potential therapeutics with established safety profiles that can be used individually or in combinations for targeting molecular regulators of replication or the survival of SARS-CoV-2. While these strategies are unlikely to provide immunity or cure, they may identify therapeutics that can alter the clinical course of COVID-19 [3-8].

The betacoronavirus genome encodes structural proteins, including the glycosylated spike (S) protein that serves as a major inducer of host immune responses. The spike protein mediates host cell invasion via binding to angiotensin converting enzyme 2 (ACE2) (a homolog of angiotensin converting enzyme ACE) which is a membrane bound carboxypeptidase). The cellular invasion process appears to be mediated by priming of the S protein facilitated by the host cell-produced serine protease TMPRSS2. In addition, the viral genome also encodes nonstructural proteins including RNA-dependent RNA polymerase (RdRp), coronavirus main protease (M^{pro}), and papain-like protease (PLpro) [5,9,10].

Therefore, targeting ACE2, TMPRSS211, RdRp, Mpro, and PLpro, as individual targets, or in combination, is a viable strategy for repurposed drugs. To that end, different drug repurposing efforts have been executed starting with data driven framework coupled with in vitro assays. One study demonstrated the potential of a poly-ADPribose polymerase 1 (PARP1) inhibitor, CVL218, currently in a Phase I clinical trial, that may serve as a potential drug candidate to inhibit SARS-CoV-2 replication in a dose-dependent manner and with no obvious cytopathic effect. CVL218 showed potential binding affinity of the N-terminal domain of nucleocapsid (N) protein of COVID-19 virus via in-silico analysis[11].

We adopted a repositioning approach using in-silico molecular modeling to screen FDA approved drugs with established safety profiles for potential inhibitory effects on COVID-19 virus. Starting with the published crystal structure provided us with structural insights for the catalytic binding domain and active druggable sites, while concurrently elucidating free binding energies with respect to binding affinity and interactions. Our structure-based approach for drug screening was focused on targeting COVID-19 virus M^{pro} based on the elegant work that resulted in solving the crystal structure of COVID-19 M^{pro} in complex with an inhibitory peptide N3 (PDB ID:6LU7). In particular, that inhibitory peptide binds the substrate-binding pocket of COVID-19 Mpro. This domain was the focus of our screen with regards to the potential hydrophobic binding domain and considering the hydrogen bond network.

We started with structure-based drug screening of more than 2000 FDA approved drugs against COVID-19 virus main M^{pro} substrate-binding pocket focusing on two potential sites (central and terminal sites) to identify potential hits based on their binding energies, binding modes, interacting amino acids, and therapeutic indications. In addition, we screened the top hits at both binding sites for potential covalent binding via nucleophilic thiol attack of Cys 145. We also elucidated the preliminary pharmacophore features for the top candidates using the three strategies bound to COVID-19 virus M^{pro} substrate-binding pocket. Finally, we performed in vitro viral replication assays on a number of the top hits in Vero cells to confirm antiviral activity.

The top hits that bound to the central site of the M^{pro} substrate-binding pocket included anti-viral drugs such as darunavir, nelfinavir and saquinavir (some of which are already being tested in COVID-19 patients,) as well as the hypercholesterolemia drug rosuvastatin and the anti-malarial drug atovaquone. The top hits that bound to the terminal site of the M^{pro} substrate-binding pocket include the anti-asthma drug montelukast and the anti-histamine drug fexofenadine, among others. Finally, the top candidates predicted to undergo covalent binding were atovaqoune, mitoxantrone, and metamizole.

We performed viral replication assays on all top hits from our in silico screen. The only 4 drugs that inhibited viral replication out of the top hits using the 3 docking strategies were all predicted to bind covalently to Cys145. These drugs are Atovaquone, Ouabain, Dronedarone, and Mebendazole. We determined the following IC50s in Vero E6 cells: Atovaquone (1.5□μM); Ouabain (0.030 μM); Mebendazole (0.25-1.2 μM) and Dronedarone (1.5 μM– however dronedarone antiviral was likely secondary to cell toxicity). Drondarone had significant cellular toxicity, while ouabain is no longer approved in the US and mebendazole in not marketed in the US any longer. Therefore, the top hit from our screen than can be immediately repurposed for treatment of COVID-19 patients is atovaquone.

Atovaquone:

Investigations into the treatment for COVID-19 span different mechanisms, as no consensus has been reached on what is the main driver of pneumonia, respiratory failure and systemic complications. One approach is antiviral in which the investigational drug would clear viral infection leading to improvement in clinical outcome; an alternative approach is anti-inflammatory in which the investigational drug would reduce levels of inflammatory cytokines to improve clinical outcomes. Yet, other mechanisms are being targeted to treat systemic complications of COVID-19 such as anti-thrombotics to prevent and treat associated venous and arterial thromboses. Which mechanism is the main driver of clinical morbidity and mortality is unknown. Currently the most promising of the antiviral therapies under investigation, remdesivir, requires IV administration, is only available in the setting of a clinical trial or by eIND mechanism for compassionate use, and cannot be used in those with severe kidney disease. The oral anti-malaria drugs hydroxychloroquine and chloroquine are being evaluated for COVID-19 treatment and also prophylaxis, but concerns have been raised because of toxicities, most notably that both drugs prolong the QT interval with reported cases of Torsades de Pointes associated with each.

In the present study, we chose to focus on the antiviral approach. We seek to evaluate atovaquone to (1) determine antiviral activity (2) to explore if viral reduction or clearance, if achieved, is associated with improvement in clinical outcome.

Atovaquone is an oral solution used as treatment of pneumocystis jirovecii pneumonia (PJP), for those unable to receive or tolerate trimethoprim-sulfamethoxazole, at a dose of

750 mg twice a day for 21 days. Atovaquone is also used for prophylaxis against PJP at a dose of 1500 mg daily. Other labeled indications include: toxoplasmosis treatment and babesiosis treatment, at doses up to 3000mg daily in 2-4 divided doses.

Mechanism of action and spectrum of activity : Atovaquone is a competitive inhibitor of ubiquinol and specifically inhibits the mitochondrial electron transport chain at the *bc1* complex[12]. Atovoquone is broad-spectrum and is used to treat malaria with an IC_{50} against malaria *in vitro* of 1-3.5nM. However, it has high levels of plasma protein binding substantially decreasing levels of unbound atovaquone[12]. Atovaquone has also been found to inhibit arboviruses such as Zika and chikungunya virus replication by depletion of intracellular nucleotides[13]. Additionally, atovaquone has been shown to reduce the interleukin-6/STAT3 signaling pathway in myeloma cells and in acute myeloid leukemia[14].

Drug concentration:

Molecular weight: 366.837 g/mol

MEPRON Suspension has been administered at dosage regimens of 500 mg once daily, 750 mg once daily, and 1,000 mg once daily to deduce C_{max} concentrations at 15.1 ± 6.1 (40.89 µM), 15.3 ± 7.6 (41.7 µM), and 16.8 ± 6.4 µg/mL (45.79 µM), respectively.

In the treatment of PJP, average ($C_{avg/ss}$) plasma concentrations of 10 to <15 µg/mL and 15 to <20µg/mL yielded 79% to 95% success in treatment. The bioavailability of atovaquone is variable as determined in 6 clinical trials in HIV patients when used for the treatment of PJP. This is likely to due to solubility and the effect of food on GI absorption. In a study of 33 patients with either HIV or hematological malignancy requiring atovaquone, patients were dosed 750 mg twice a day and C_{min} was obtained 12 hours after evening dose and C_{max} 1-5 hours after morning dose. The median C_{min} (IQR) was 11.3 µg/ml (6.2-27.8) and median C_{max} (IQR) was 13.4 µg/mL (6.0-28.3) and were not different between HIV and non-HIV patients. Nineteen of 33 (58%) had C_{min} <15µg/mL which may impact clinical response for PJP.

Metabolism: Under normal conditions, there is no evidence that atovaquone is significantly metabolized in humans, or that metabolism is required for drug elimination. It may be possible that certain enzymes could be induced and therefore lead to increased atovaquone biotransformation, but this has not been demonstrated [15].

Elimination: The pharmacokinetic half-life of atovaquone was 62.5 ± 35.3 hours after IV administration and ranges from 67.0 ± 33.4 to 77.6 ± 23.1 hours across studies following administration of MEPRON Suspension. Atovaquone pharmacokinetics are characterized by an extremely long elimination half-life of 50–84 h after oral administration of 250 mg [16].

Distribution: Atovaquone is extensively bound to plasma proteins (99.9%) and therefore the concentration of unbound atovaquone in the vascular compartment will be significantly lower than measured plasma concentrations[17]. In a study of atovaquone population pharmacokinetics, the volume of distribution of atovaquone was 7.98 L/kg, although individual values were markedly linked to body weight; the volume of distribution shows a linear increase with increased patient body weight[18]. This volume of distribution indicates that atovaquone is not confined to the intravascular space.

We were unable to find literature describing the tissue concentration of atovaquone following systemic administration, however, several reports suggest that atovaquone is not confined to the intravascular space, despite being highly bound to albumin given that it has a low drug clearance rate, which suggests that it likely accumulates in tissue[12]

Another level of evidence in support of tissue accumulation of atovaquone is the effective concentration needed for its therapeutic effect. While the IC_{50} for atovaquone against falciparum malaria is in the low nanomolar range, the IC_{50} for atovaquone for pneumocystis jirovecii pneumonia is from **0.1-3.0 ug/ml, which is equivalent to 8.178 μM.**

The IC_{50} for atovaquone against SARS-CoV-2 in our assay is approximately 800 nM, which is well within the therapeutic range as outlined above. Therefore, we predict that the inhibitory effect on SARS-CoV-2 is well within the FDA approved dosing regimen.

Drug interaction: Atovaquone exposure is markedly decreased when taken with rifampin.

Tolerability: Atovaquone has been found to be generally well tolerated. Adverse events have been mild and include rash, fever, diarrhea, abdominal pain, and headache.

Current studies in COVID-19: Currently, an independent single center, single arm open-label trial is underway examining atovaquone 750 mg bid for up to 10 days in combination with azithromycin 500mg X 1 followed by 250mg daily for up to 10 days for COVID-19. [NCT04339426] However, this study is limited by evaluation of atovaquone only in combination with azithromycin and by the absence of a contemporary control group for comparison.

Design of the ATaQ COVID-19 Trial

The purpose of the current study is to accelerate the use of a clinically available therapeutic already FDA-approved for other indications in the setting of pandemic COVID-19 addressing a serious and emergent unmet medical need. In consideration of the information included in this protocol, the overall risks to participants are outweighed by

the potential benefits of atovaquone experimental therapy for the treatment of COVID-19. The benefit-risk balance for this study is considered positive.

Inclusion Criteria:

1. Diagnosis of COVID-19 by positive RT-PCR requiring hospitalization within 72 hours
2. Age ≥18 years old
3. Able to provide informed consent, or (as allowed by IRB), immediate availability of designated legally authorized representative to provide consent by proxy
4. Anticipated hospitalization for >48 hours

Exclusion Criteria

Patients who meet *any* of the following exclusion criteria are not to be enrolled in this study:

1) Participation in any other clinical trial with antiviral activity against COVID-19
2) Breastfeeding women
3) Known hypersensitivity to atovaquone or formulation excipient

4) Active treatment with rifampin

5) HIV patients with AIDS requiring treatment for *Pneumocystis jirovecii* or *Toxoplasma gondii*

6) Not expected to survive for 72 hours.

7) >14 days from symptom onset

Randomization

Patients who meet eligibility criteria and volunteer to participate will be randomized in a 2:1 ratio to atovaquone or placebo on Day 1 using computerized randomization. An unblinded investigational pharmacist not otherwise involved in the trial will know treatment assignment and dispense investigational product. As GI absorption of atovaquone increased when taken with food, so we will administer with a meal or snack.

Blinding

Double blinding of treatment assignments will be performed in this study, with the study team and patients blinded to treatment assignment.

The list of concomitant medications will be assessed only from Day 1 prior to enrollment to Day 15 or discharge, whichever is earlier.

Patient Enrollment and Treatment Assignment

Entry into screening does not guarantee enrollment into the study. In order to manage the total study enrollment, the study researchers may suspend screening and/or enrollment at any at any time.

Pretreatment Assessments

Screening Visit

Patients will be screened within 2 days before randomization and dosing to determine eligibility for participation in the study. Screening will occur under approved HIPAA waiver for research to identify and screen all hospitalized COVID-19 positive patients on a daily basis.

Obtain informed consent.

After informed consent has been negotiated and the form signed, the following assessments will be performed to determine eligibility requirements as specified in the inclusion and exclusion criteria:

- Review of focused medical history including the following information (e.g., date of first symptoms, overall symptoms, exposure source, demographics, baseline characteristics), allergies and past medical history.
- Review and record medications and therapies for the current illness
- Recording of vital signs (heart rate, temperature, blood pressure), body weight, and height
- Documentation of respiratory support: Respiratory Rate, Oxygen supplementation: room air, nasal canula, face mask, non-rebreather, high-flow device, mechanical ventilation; and FiO_2
- SpO_2 at rest or PaO2
- Radiographic findings

Study patients who qualify and volunteer to participate should be immediately consented and randomized. Randomization and initiation of dosing should occur on the same day if possible.

Baseline/Day 1 Assessments

The following evaluations are to be completed at the Day 1 visit. The investigator must have confirmed eligibility and signing of consent before proceeding with randomization on the Day 1 visit, followed immediately by first dose of investigational product. The assessments can be completed by the patient care team and do not need to be repeated by research personnel. The following assessments must be documented before administering investigational product, using the most recent data available at the time of randomization:

Recording of vital signs (heart rate, temperature, blood pressure, body weight, height)

Documentation of respiratory status:

Respiratory rate

Oxygen supplementation and FiO_2: room air, nasal canula, face mask, non-rebreather, noninvasive ventilation or high flow oxygen devices, mechanical ventilation, or ECMO

Oxygenation: (SpO_2 or PaO_2)

Radiographic findings (if available)

Review AEs and document concomitant medications

Document Ordinal Scale at baseline

Obtain saliva sample and nasopharyngeal swab sample for viral load quantification at day 1 prior to initial dose

Obtain blood for research sample

Daily Study Assessments (Days 2-10)

The following evaluations are to be documented daily from Days 2 – 10 or until discharge whichever comes earlier, using the data recorded at or closest to 12:00 noon each day:

- Vital signs (heart rate, temperature, blood pressure), body weight (if available).
- Documentation of respiratory status: Respiratory rate, Oxygen supplementation and FiO_2: room air, nasal canula, face mask, non-rebreather, noninvasive ventilation or high flow oxygen devices, mechanical ventilation, or ECMO
- Oxygenation: (SpO_2 or PaO_2)
- Radiographic findings (if available)
- Review of AEs and document concomitant medications
- Saliva sample for COVID-19 RT-PCR every 12 hours (Days 2-8)
- Saliva sample for COVID-19 RT-PCR once daily (Days 9-10)
- Additional blood draws for biobanking (Day 3, and 5 only)
- A patient who does not have 5 saliva samples collected prior to discharge, the patient may be given saliva kits for home collection if deemed that patient can follow instructions and collect saliva correctly.

Clinical Laboratory Assessments

Clinical laboratory assessments will be conducted as clinically indicated and all laboratory testing will be completed by local laboratories. Clinical laboratory data to be captured in the trial database will include serum chemistries, liver function tests, complete blood counts including absolute neutrophil count, CRP, hs-CRP, D-dimer, ferritin, IL-6, troponin, NTpBNP.

SARS-CoV-2 testing will include RT-qPCR to detect or quantify SARS-CoV-2 or virus sequencing results from saliva (baseline and daily until discharge or death, and 8 days and last day of hospitalization or Day 15 if still hospitalized.

Pretreatment and posttreatment samples with detectable SARS-CoV-2 may be sequenced for resistance monitoring of the viral polymerase gene. For all clinical laboratory tests, except those at Day 1, when more than 1 result is available in a calendar

day, the value closest to 12:00 noon should be captured in the eCRF. For Day 1 tests, the most recent result before dosing should be used.

Physical Examination

No physical examination is mandated by the study protocol beyond the capture of vital signs (heart rate, respiratory rate, temperature, blood pressure, SpO_2 at rest or PaO_2) as documented clinically.

Post-treatment Assessments

Treatment will continue to complete a 10 Day course or until viral clearance is documented, whichever occurs first.

Telephone call on Day 15 and 29 for those discharged. The phone call will include a brief survey on symptoms and information on any re-hospitalizations.

Final review of AEs and concomitant medication

Vital signs will be captured if still inpatient and the ordinal scale will be assessed.

Assessments for Early Discontinuation from Study

If a patient discontinues study dosing (for example, as a result of an AE, intolerance of investigational product, or clinically significant laboratory abnormality felt to be at least possibly trial-related), every attempt should be made to keep the patient in the study and continue to perform the required study-related follow-up and procedures. If this is not possible or acceptable to the patient or investigator, the patient may be discontinued from the trial, with phone call for vital status on trial Day 29.

Drug Dosage Reduction:

Atovaquone should be given with food to increase absorption and to possibly limit GI intolerance. Known adverse events including nausea, vomiting and diarrhea. If patient develops moderate to severe GI symptoms, a reduction in dose to 750 mg bid can be instituted per investigator discretion to enhance the likelihood for the patient to complete therapy.

Criteria for Discontinuation of Study Treatment

Study medication may be interrupted or discontinued in the following instances:

- Intercurrent illness that would, in the judgment of the investigator, affect assessments of clinical status to a significant degree. Following resolution of intercurrent illness, the patient may resume study dosing at the discretion of the investigator.
- Unacceptable toxicity, or toxicity that, in the judgment of the investigator, compromises the ability to continue study-specific procedures or is considered to not be in the patient's best interest

- Patient request to discontinue for any reason
- Discontinuation of the study at the request of a regulatory agency or an institutional review board (IRB)
- DSMB recommends termination of trial
- Patient may be able to receive other treatments for COVID-19 at the discretion of the treating providers if there is disease progression or after completion of 10 days of treatment. Study medication does not need to be stopped when additional medication is started.

End of Study

The end of the study will occur when the last participant's last observation (or visit).

Post Study Care

The long-term care of the participant will remain the responsibility of their primary treating provider. Atovaquone is being supplied with curative intent. There is no provision for post-study availability.

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ATAQ Trial Statistical Analysis Plan

Prospectively will omit any sample with no detectable GADPH housekeeping and any with GADPH Ct >34

Values below detection limit will be assigned a value ½ between lowest detection limit and zero

All analyses will be intention-to-treat

Primary analysis: Between group differences in viral load (Log copy number/ml) using generalized linear mixed-effect models of repeated measures (GLMM), using data from all samples

Secondary/exploratory analyses:

1. Between group differences in viral load (Log copy number/ml) using GLMM, assessing differences at at 3, 5, and 7 days
2. AUC comparison between groups of viral load (Log copy number/ml) through day 3 and day 7 via the trapezoidal rule.
3. Between group differences in viral load (Log copy number/ml) using GLMM stratified by
    a. Morning (a) and evening (b) samples
    b. By any remdesivir use
    c. By any monoclonal or polyclonal antibody use
    d. By median split of baseline values
    e. By median split time from onset of symptoms
    f. By median split of BMI
    g. By DM status
    h. Sex
    i. Age
4. Between group comparison of time to drop in viral load (Log copy number/ml) of 2 log units using Kaplan-Meier estimation
5. Change of ≥2 points on the ordinal scale at Day 5 by chi-square analysis